

LAP20 Res'd PCT/PTO 10 FEB 2006

<110> Sirna Therapeutics, Inc.
Radka, Susan
Zinnen, Shawn
Jadhavi, Vasant
McSwiggen, James
Vaish, Narendra

<120> Detection and Quantitation of Nucleic Acid Molecules in
Biological Samples

<130> 700/020 (MBHB03-770-B)

<140> PCT/US 04/026857
<141> 2004-08-17

<150> US 60/497,222
<151> 2003-08-22

<150> US 60/543,784
<151> 2004-02-11

<160> 24

<170> PatentIn version 3.3

<210> 1
<211> 21
<212> RNA
<213> Artificial Sequence

<220>
<223> Description of Artificial Sequence: siNA sense region

<220>
<221> misc_feature
<222> (1)..(1)
<223> 5'-3' attached terminal deoxyabasic moiety

<220>
<221> misc_feature
<222> (1)..(1)
<223> 2'-deoxy-2'-fluoro

<220>
<221> misc_feature
<222> (3)..(3)
<223> 2'-deoxy-2'-fluoro

<220>
<221> misc_feature
<222> (5)..(5)
<223> 2'-deoxy-2'-fluoro

<220>
<221> misc_feature
<222> (7)..(10)
<223> 2'-deoxy-2'-fluoro

<220>
<221> misc_feature
<222> (12)..(15)
<223> 2'-deoxy-2'-fluoro

<220>

<221> misc_feature
 <222> (17)..(19)
 <223> 2'-deoxy-2'-fluoro

<220>
 <221> misc_feature
 <222> (20)..(20)
 <223> n stands for thymidine

<220>
 <221> misc_feature
 <222> (21)..(21)
 <223> 3'-3 attached terminal deoxyabasic moiety

<400> 1
 ugugcacuuc gcuucaccun n

21

<210> 2
 <211> 23
 <212> RNA
 <213> Artificial Sequence

<220>
 <223> Description of Artificial Sequence: Chol-7/siNA sense region

<220>
 <221> misc_feature
 <222> (1)..(1)
 <223> n stands for thymidine with 5'bifunctional cholesterol conjugate

<220>
 <221> misc_feature
 <222> (3)..(3)
 <223> inverted deoxyabasic moiety

<220>
 <221> misc_feature
 <222> (3)..(3)
 <223> 2'-deoxy-2'-fluoro

<220>
 <221> misc_feature
 <222> (5)..(5)
 <223> 2'-deoxy-2'-fluoro

<220>
 <221> misc_feature
 <222> (7)..(7)
 <223> 2'-deoxy-2'-fluoro

<220>
 <221> misc_feature
 <222> (9)..(12)
 <223> 2'-deoxy-2'-fluoro

<220>
 <221> misc_feature
 <222> (14)..(17)
 <223> 2'-deoxy-2'-fluoro

<220>
 <221> misc_feature
 <222> (19)..(21)
 <223> 2'-deoxy-2'-fluoro

<220>
 <221> misc_feature
 <222> (22)..(23)
 <223> n stands for thymidine

<220>
 <221> misc_feature
 <222> (23)..(23)
 <223> 3'-3 attached terminal deoxyabasic moiety

<400> 2
 naugugcacu ucgcuucacc unn

23

<210> 3
 <211> 27
 <212> RNA
 <213> Artificial Sequence

<220>
 <223> Description of Artificial Sequence: Chol-trigal 7/siNA sense region

<220>
 <221> misc_feature
 <222> (1)..(1)
 <223> n stands for thymidine with 5'bifunctional cholesterol conjugate

<220>
 <221> misc_feature
 <222> (3)..(3)
 <223> inverted deoxyabasic moiety

<220>
 <221> misc_feature
 <222> (3)..(3)
 <223> 2'-deoxy-2'-fluoro

<220>
 <221> misc_feature
 <222> (5)..(5)
 <223> 2'-deoxy-2'-fluoro

<220>
 <221> misc_feature
 <222> (7)..(7)
 <223> 2'-deoxy-2'-fluoro

<220>
 <221> misc_feature
 <222> (9)..(12)
 <223> 2'-deoxy-2'-fluoro

<220>
 <221> misc_feature
 <222> (14)..(17)
 <223> 2'-deoxy-2'-fluoro

<220>
 <221> misc_feature
 <222> (19)..(21)
 <223> 2'-deoxy-2'-fluoro

<220>
 <221> misc_feature
 <222> (22)..(23)
 <223> n stands for thymidine

<220>
 <221> misc_feature
 <222> (24)..(24)
 <223> inverted deoxyabasic moiety

<220>
 <221> misc_feature
 <222> (25)..(25)
 <223> 2'-deoxy-2'-fluoro

<220>
 <221> misc_feature
 <222> (26)..(26)
 <223> n stands for thymidine

<220>
 <221> misc_feature
 <222> (27)..(27)
 <223> n stands for tri-N-acetyl galactosamine conjugate

<220>
 <221> misc_feature
 <222> (27)..(27)
 <223> 3'-3' attached terminal deoxyabasic moiety

<400> 3
 naugucacu ucgcucacc unnnann

27

<210> 4
 <211> 21
 <212> RNA
 <213> Artificial Sequence

<220>
 <223> Description of Artificial Sequence: siNA antisense region

<220>
 <221> misc_feature
 <222> (1)..(3)
 <223> 2'-O-methyl

<220>
 <221> misc_feature
 <222> (4)..(4)
 <223> 2'-deoxy-2'-fluoro

<220>
 <221> misc_feature
 <222> (5)..(8)
 <223> 2'-O-methyl

<220>
 <221> misc_feature
 <222> (9)..(9)
 <223> 2'-deoxy-2'-fluoro

<220>
 <221> misc_feature
 <222> (10)..(13)
 <223> 2'-O-methyl

<220>
 <221> misc_feature
 <222> (14)..(14)

<223> 2'-deoxy-2'-fluoro

<220>

<221> misc_feature

<222> (15)..(15)

<223> 2'-O-methyl

<220>

<221> misc_feature

<222> (16)..(16)

<223> 2'-deoxy-2'-fluoro

<220>

<221> misc_feature

<222> (17)..(17)

<223> 2'-O-methyl

<220>

<221> misc_feature

<222> (18)..(18)

<223> 2'-deoxy-2'-fluoro

<220>

<221> misc_feature

<222> (19)..(19)

<223> 2'-O-methyl

<220>

<221> misc_feature

<222> (20)..(20)

<223> Phosphorothioate 3'-Internucleotide Linkage

<220>

<221> misc_feature

<222> (21)..(21)

<223> n stands for thymidine

<400> 4

aggugaagcg aagugcacan n

21

<210> 5

<211> 10

<212> RNA

<213> Artificial Sequence

<220>

<223> Description of Artificial Sequence: Biotinyl capture oligo/siNA sense region

<220>

<221> misc_feature

<222> (1)..(1)

<223> 5' attached terminal Biotin

<220>

<221> misc_feature

<222> (1)..(10)

<223> 2'-O-methyl

<400> 5

aggugaagcg

10

<210> 6

<211> 10

<212> RNA
 <213> Artificial Sequence

<220>
 <223> Description of Artificial Sequence: Biotinyl capture oligo/siNA antisense region

<220>
 <221> misc_feature
 <222> (1)..(1)
 <223> 5'attached terminal Biotin

<220>
 <221> misc_feature
 <222> (1)..(10)
 <223> 2'-O-methyl

<400> 6
 ugugcacuuc 10

<210> 7
 <211> 11
 <212> RNA
 <213> Artificial Sequence

<220>
 <223> Description of Artificial Sequence: FITC labeled reporter oligo/siNA sense region

<220>
 <221> misc_feature
 <222> (1)..(11)
 <223> 2'-O-methyl

<220>
 <221> misc_feature
 <222> (11)..(11)
 <223> 3' attached terminal Fluorescein

<400> 7
 aagugcacau u 11

<210> 8
 <211> 11
 <212> RNA
 <213> Artificial Sequence

<220>
 <223> Description of Artificial Sequence: FITC labeled reporter oligo/siNA antisense region

<220>
 <221> misc_feature
 <222> (1)..(11)
 <223> 2'-O-methyl

<220>
 <221> misc_feature
 <222> (11)..(11)
 <223> 3' attached terminal Fluorescein

<400> 8

gcuucacuu u

<210> 9
 <211> 20
 <212> RNA
 <213> Artificial Sequence

<220>
 <223> Description of Artificial Sequence: siNA sense region

<220>
 <221> misc_feature
 <222> (2)..(2)
 <223> 2'-deoxy-2'-fluoro

<220>
 <221> misc_feature
 <222> (4)..(4)
 <223> 2'-deoxy-2'-fluoro

<220>
 <221> misc_feature
 <222> (6)..(9)
 <223> 2'-deoxy-2'-fluoro

<220>
 <221> misc_feature
 <222> (11)..(14)
 <223> 2'-deoxy-2'-fluoro

<220>
 <221> misc_feature
 <222> (16)..(18)
 <223> 2'-deoxy-2'-fluoro

<220>
 <221> misc_feature
 <222> (19)..(19)
 <223> n stands for thymidine

<220>
 <221> misc_feature
 <222> (20)..(20)
 <223> 3'-3' attached terminal deoxyabasic moiety

<400> 9
 gugcacuucg cuucaccunn

<210> 10
 <211> 19
 <212> RNA
 <213> Artificial Sequence

<220>
 <223> Description of Artificial Sequence: siNA sense region

<220>
 <221> misc_feature
 <222> (1)..(1)
 <223> 2'-deoxy-2'-fluoro

<220>
 <221> misc_feature

<222> (3)..(3)
 <223> 2'-deoxy-2'-fluoro

<220>
 <221> misc_feature
 <222> (5)..(8)
 <223> 2'-deoxy-2'-fluoro

<220>
 <221> misc_feature
 <222> (10)..(13)
 <223> 2'-deoxy-2'-fluoro

<220>
 <221> misc_feature
 <222> (15)..(17)
 <223> 2'-deoxy-2'-fluoro

<220>
 <221> misc_feature
 <222> (18)..(18)
 <223> n stands for thymidine

<220>
 <221> misc_feature
 <222> (19)..(19)
 <223> 3'-3 attached terminal deoxyabasic moiety

<400> 10
 ugcacuucgc uucaccunn

19

<210> 11
 <211> 17
 <212> RNA
 <213> Artificial Sequence

<220>
 <223> Description of Artificial Sequence: siNA sense region

<220>
 <221> misc_feature
 <222> (1)..(1)
 <223> 2'-deoxy-2'-fluoro

<220>
 <221> misc_feature
 <222> (3)..(6)
 <223> 2'-deoxy-2'-fluoro

<220>
 <221> misc_feature
 <222> (8)..(11)
 <223> 2'-deoxy-2'-fluoro

<220>
 <221> misc_feature
 <222> (13)..(15)
 <223> 2'-deoxy-2'-fluoro

<220>
 <221> misc_feature
 <222> (16)..(16)
 <223> n stands for thymidine

<220>

<221> misc_feature
 <222> (17)..(17)
 <223> 3'-3 attached terminal deoxyabasic moiety

<400> 11
 cacuucgcuu caccunn

17

<210> 12
 <211> 13
 <212> RNA
 <213> Artificial Sequence

<220>
 <223> Description of Artificial Sequence: siNA sense region

<220>
 <221> misc_feature
 <222> (1)..(2)
 <223> 2'-deoxy-2'-fluoro

<220>
 <221> misc_feature
 <222> (4)..(7)
 <223> 2'-deoxy-2'-fluoro

<220>
 <221> misc_feature
 <222> (9)..(11)
 <223> 2'-deoxy-2'-fluoro

<220>
 <221> misc_feature
 <222> (12)..(12)
 <223> n stands for thymidine

<220>
 <221> misc_feature
 <222> (13)..(13)
 <223> 3'-3 attached terminal deoxyabasic moiety

<400> 12
 ucgcuucacc unn

13

<210> 13
 <211> 20
 <212> RNA
 <213> Artificial Sequence

<220>
 <223> Description of Artificial Sequence: siNA antisense region

<220>
 <221> misc_feature
 <222> (3)..(3)
 <223> 2'-deoxy-2'-fluoro

<220>
 <221> misc_feature
 <222> (8)..(8)
 <223> 2'-deoxy-2'-fluoro

<220>
 <221> misc_feature

<222> (13)..(13)
<223> 2'-deoxy-2'-fluoro

<220>
<221> misc_feature
<222> (15)..(15)
<223> 2'-deoxy-2'-fluoro

<220>
<221> misc_feature
<222> (17)..(17)
<223> 2'-deoxy-2'-fluoro

<220>
<221> misc_feature
<222> (19)..(19)
<223> Phosphorothioate 3'-Internucleotide Linkage

<220>
<221> misc_feature
<222> (20)..(20)
<223> n stands for thymidine

<400> 13
ggugaagcga agugcacann

20

<210> 14
<211> 19
<212> RNA
<213> Artificial Sequence

<220>
<223> Description of Artificial Sequence: siNA antisense region

<220>
<221> misc_feature
<222> (2)..(2)
<223> 2'-deoxy-2'-fluoro

<220>
<221> misc_feature
<222> (7)..(7)
<223> 2'-deoxy-2'-fluoro

<220>
<221> misc_feature
<222> (12)..(12)
<223> 2'-deoxy-2'-fluoro

<220>
<221> misc_feature
<222> (14)..(14)
<223> 2'-deoxy-2'-fluoro

<220>
<221> misc_feature
<222> (16)..(16)
<223> 2'-deoxy-2'-fluoro

<220>
<221> misc_feature
<222> (18)..(18)
<223> Phosphorothioate 3'-Internucleotide Linkage

<220>

<221> misc_feature
 <222> (19)..(19)
 <223> n stands for thymidine

<400> 14
 gugaagcgaa gugcacann

19

<210> 15
 <211> 17
 <212> RNA
 <213> Artificial Sequence

<220>
 <223> Description of Artificial Sequence: siNA antisense region

<220>
 <221> misc_feature
 <222> (5)..(5)
 <223> 2'-deoxy-2'-fluoro

<220>
 <221> misc_feature
 <222> (10)..(10)
 <223> 2'-deoxy-2'-fluoro

<220>
 <221> misc_feature
 <222> (12)..(12)
 <223> 2'-deoxy-2'-fluoro

<220>
 <221> misc_feature
 <222> (14)..(14)
 <223> 2'-deoxy-2'-fluoro

<220>
 <221> misc_feature
 <222> (16)..(16)
 <223> Phosphorothioate 3'-Internucleotide Linkage

<220>
 <221> misc_feature
 <222> (17)..(17)
 <223> n stands for thymidine

<400> 15
 gaagcgaagu gcacann

17

<210> 16
 <211> 15
 <212> RNA
 <213> Artificial Sequence

<220>
 <223> Description of Artificial Sequence: siNA antisense region

<220>
 <221> misc_feature
 <222> (3)..(3)
 <223> 2'-deoxy-2'-fluoro

<220>
 <221> misc_feature

<222> (8)..(8)
 <223> 2'-deoxy-2'-fluoro

<220>
 <221> misc_feature
 <222> (10)..(10)
 <223> 2'-deoxy-2'-fluoro

<220>
 <221> misc_feature
 <222> (12)..(12)
 <223> 2'-deoxy-2'-fluoro

<220>
 <221> misc_feature
 <222> (14)..(14)
 <223> Phosphorothioate 3'-Internucleotide Linkage

<220>
 <221> misc_feature
 <222> (15)..(15)
 <223> n stands for thymidine

<400> 16
 agcgaagugc acann

15

<210> 17
 <211> 21
 <212> RNA
 <213> Artificial Sequence

<220>
 <223> Description of Artificial Sequence: siNA sense region

<220>
 <221> misc_feature
 <222> (1)..(1)
 <223> 5'-3' attached terminal deoxyabasic moiety

<220>
 <221> misc_feature
 <222> (1)..(1)
 <223> 2'-deoxy-2'-fluoro

<220>
 <221> misc_feature
 <222> (2)..(2)
 <223> 2'-deoxy

<220>
 <221> misc_feature
 <222> (3)..(3)
 <223> 2'-deoxy-2'-fluoro

<220>
 <221> misc_feature
 <222> (4)..(4)
 <223> 2'-deoxy

<220>
 <221> misc_feature
 <222> (5)..(5)
 <223> 2'-deoxy-2'-fluoro

<220>

<221> misc_feature
<222> (6)..(6)
<223> 2'-deoxy

<220>
<221> misc_feature
<222> (7)..(10)
<223> 2'-deoxy-2'-fluoro

<220>
<221> misc_feature
<222> (11)..(11)
<223> 2'-deoxy

<220>
<221> misc_feature
<222> (12)..(15)
<223> 2'-deoxy-2'-fluoro

<220>
<221> misc_feature
<222> (16)..(16)
<223> 2'-deoxy

<220>
<221> misc_feature
<222> (17)..(19)
<223> 2'-deoxy-2'-fluoro

<220>
<221> misc_feature
<222> (20)..(20)
<223> n stands for thymidine

<220>
<221> misc_feature
<222> (21)..(21)
<223> 3'-3 attached terminal deoxyabasic moiety

<400> 17
ugugcacuuc gcuucaccun n

21

<210> 18
<211> 21
<212> RNA
<213> Artificial Sequence

<220>
<223> Description of Artificial Sequence: siNA antisense region

<220>
<221> misc_feature
<222> (1)..(3)
<223> 2'-O-methyl

<220>
<221> misc_feature
<222> (4)..(4)
<223> 2'-deoxy-2'-fluoro

<220>
<221> misc_feature
<222> (5)..(8)
<223> 2'-O-methyl

<220>
<221> misc_feature
<222> (9)..(9)
<223> 2'-deoxy-2'-fluoro

<220>
<221> misc_feature
<222> (10)..(13)
<223> 2'-O-methyl

<220>
<221> misc_feature
<222> (14)..(14)
<223> 2'-deoxy-2'-fluoro

<220>
<221> misc_feature
<222> (15)..(15)
<223> 2'-O-methyl

<220>
<221> misc_feature
<222> (16)..(16)
<223> 2'-deoxy-2'-fluoro

<220>
<221> misc_feature
<222> (17)..(17)
<223> 2'-O-methyl

<220>
<221> misc_feature
<222> (18)..(18)
<223> 2'-deoxy-2'-fluoro

<220>
<221> misc_feature
<222> (19)..(19)
<223> 2'-O-methyl

<220>
<221> misc_feature
<222> (20)..(20)
<223> Phosphorothioate 3'-Internucleotide Linkage

<220>
<221> misc_feature
<222> (21)..(21)
<223> n stands for thymidine

<400> 18
aggugaagcg aagugcacan n

21

<210> 19
<211> 21
<212> RNA
<213> Artificial Sequence

<220>
<223> Description of Artificial Sequence: siNA sense region

<220>
<221> misc_feature
<222> (1)..(1)
<223> 5'-3 attached terminal deoxyabasic moiety

<220>
 <221> misc_feature
 <222> (20)..(20)
 <223> n stands for thymidine

<220>
 <221> misc_feature
 <222> (21)..(21)
 <223> 3'-3 attached terminal deoxyabasic moiety

<400> 19
 cugaguuuaa aaggcaccn n

21

<210> 20
 <211> 21
 <212> RNA
 <213> Artificial Sequence

<220>
 <223> Description of Artificial Sequence: siNA antisense region

<220>
 <221> misc_feature
 <222> (20)..(20)
 <223> Phosphorothioate 3'-Internucleotide Linkage

<220>
 <221> misc_feature
 <222> (21)..(21)
 <223> n stands for thymidine

<400> 20
 gggugccuuu uaaacucagn n

21

<210> 21
 <211> 13
 <212> RNA
 <213> Artificial Sequence

<220>
 <223> Description of Artificial Sequence: Capture probe/siNA sense region

<220>
 <221> misc_feature
 <222> (1)..(13)
 <223> 2'-O-methyl

<220>
 <221> misc_feature
 <222> (13)..(13)
 <223> 3' attached terminal Biotin

<400> 21
 cuuuuaaacu cag

13

<210> 22
 <211> 9
 <212> RNA
 <213> Artificial Sequence

<220>
 <223> Description of Artificial Sequence: Reporter probe/siNA sense region

<220>
 <221> misc_feature
 <222> (1)..(1)
 <223> 5' attached terminal Fluorescein

<220>
 <221> misc_feature
 <222> (1)..(9)
 <223> 2'-O-methyl

<400> 22
 aagggugcc

9

<210> 23
 <211> 14
 <212> RNA
 <213> Artificial Sequence

<220>
 <223> Description of Artificial Sequence: Capture probe/siNA antisense region

<220>
 <221> misc_feature
 <222> (1)..(1)
 <223> 5' attached terminal Biotin

<220>
 <221> misc_feature
 <222> (1)..(14)
 <223> 2'-O-methyl

<400> 23
 uacugaguuu aaaa

14

<210> 24
 <211> 8
 <212> RNA
 <213> Artificial Sequence

<220>
 <223> Description of Artificial Sequence: Reporter probe/siNA antisense region

<220>
 <221> misc_feature
 <222> (1)..(8)
 <223> 2'-O-methyl

<220>
 <221> misc_feature
 <222> (8)..(8)
 <223> 3' attached terminal Fluorescein

<400> 24
 ggcaccca

8